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Research Article

Anthelmentic Resistance of Ovine Fasciolosis at Small Holder Farmers in Debre Birhan District, North Shawo Zone, Amhara Region Central Ethiopia

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- Fasciolosis
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- Intermediate host

Abstract

Filed experimental trial was conducted; starting from December to April 2013 for five months in Debre Birhan districts, North Showa zone, Amhara Region Central Ethiopia, to evaluate effectiveness of the anthelmintic drug. A field experiment study was used, 40 sheep selected and divided into four groups. The fecal sample was collected before as well as after treated and to examine by Modified McMaster method. The data was managed by descriptive statistics of SPSS, one way ANOVAs. The faecal egg count reduction test was commonly used for anthelmintics drugs resistance. The overall result, the efficiency of the tested anthelmintic was 81.4% triclabendazole, 63.2% albendazole and 38.2% tetraclozana; this indicates the occurrence of drug resistance. Significance difference ($p<0.05$) between control and treated groups. Efficiency of the tested anthelmintics drugs below the standard, therefore Fasciola infection developed resistance in the study area. However, among the tested anthelmintics Triclabendazole was more effective to treated Fasciolosis. One way ANOVAs result indicted that sum of squares & mean squares of Fasciola faecal egg count among treated and non-treated groups was statistically significantly ($p=0.000$) but among non-treated group difference was no statistically significance ($p=0.926$).

INTRODUCTION

Background and Justifications

Ethiopia has been large number of livestock populations in Africa, its accounting 59.5 million cattle, 30.70 million sheep, and 30.20 million goats. Ethiopia has the largest numbers of small ruminant population [1]. Additional, in Ethiopia rational prophylactic program has sound Fasciolosis control strategically programs is needed based on the local epidemiological information the parasite [2]. Apparently, losses the infestation parasitic including Fasciolosis is predictable highly in temperate area such as Ethiopia was strategic and the most effective disease control strategic program missing [3]. Flukicides are treatment liver fluke infection are triclabendazole, albendazole, tetraclozan and benzimidazoles [4]. Triclabendazole is option for treatment of ovine Fasciola, it was effectively treated both juvenile and adult liver flukes [5].

Anthelmintics resistance is expressed ability of parasite population or individual worm within population the tolerance of flukicids [6,7]. Anthelmintics drug resistance is widespread problem treatment of liver fluke infestation in sheep and goat, also almost every region of the countries and the mechanisms of action anthelmintics resistance are intrinsically related to the mode of action drugs against helminthes SPP and ability of the parasite to overcome drug action [7]. The resistance of Fasciola to flukicides was reported in Australia [5]. Significant difference of the flukicides resistance is raised regarding evaluation flukicides efficacy [8,9]. Fasciolosis is not treated with flukicides because that differ in chemical and mode of action but efficiency of flukicides against different developmental stage of the liver fluke common treated by using triclabendazole, tetraclozan and albendazole [4].

Methods of anthelmintics resistance test can be classified as *in vivo* and *in vitro* methods and have been extensively reviewed elsewhere [10]. Controlled efficacy test was considered a gold standard and reliable method to assess efficacy of antiparasitic [10]. For the diagnose anthelmintics resistance the method involves anthelmintic treated naturally infected animals otherwise, experimentally infected animals, and additional to performing the postmortem inspection when the worm recovery and for detection of the surviving or resistant worm [10]. Most conventional method is *in vivo* methods, which includes the faecal egg count reduction test plus controlled efficiency test [11]. Anthelmintics resistance was estimated that, when the efficacy was greater than 95% and with >90 lower confidence limit [12].

In vivo test involves, FECRT was a clinical trial that estimates the efficacy of an antiparasitic against helminthes infection by comparing the parasite faecal egg counts of treated animals pretreatment and post treatment FEC between treated and untreated groups [12]. Due to its feasibility and relatively lower cost most commonly used technique finding of anthelmintics resistance in field and farms [13]. Therefore, the objectives this research thesis was initiated with the aim to evaluate the efficacy of tested anthelmintics drugs against Fasciola species in study area at Debre Birhan district.

MATERIALS AND METHODS

Description of the Area

This study was conducted selected sites of the study area in Debre Birhan districts, it located at central high land of Ethiopia, in North Showa Zone, at Amhara Regional State and situated at distance of 125Km Northeast Addis Ababa, Ethiopia lying between 9°41'-9.683° N latitude and 39°32'-39°.533° E longitude and an elevation of 2,840 m.a.s.l [14]. The rainfall pattern in the study area was bimodal. The mean annual rainfall was 927.10mm Hg and its rainfall pattern is characterized by the maximum 1293.02mm Hg in August as well as the minimum 4.72mm in December. Similarly, maximum temperature of the area was 18.6°C and minimum temperature 8.20^{0}C and land coverage is 18,081.95Km^2.

Study Population

An estimated 20,200 sheep was reared around the study area in Debre Birhan districts in North Showa Zone Amhara administrative Regional State [15]. The study animal comprised different breeds indigenous breed "*Menze*", cross breed "*Menze* with *Awassi*" and "*Awassi* "breed, indigenous breed ("*Menze*") cross breed as well as *Awassi* breed, and also different age, sex, body conditions groups and category found under the extensive free grazing system in the study area [1].

Study Design

Field experimental trial: Field experimental study design was conducted to evaluate the efficacy of anthelmintics (triclabendazole, albendazole and tetraclozan) Fasciolosis in sheep, and experimental trial 40 positive sheep were selected based on the fecal egg load, which has been greater than or equal to ≧150 EPG. The animal was allocated and classifieds in to four experimental groups depends upon its sex, breed and body condition, and each experimental group with 10 sheep, group one treated with albendazole, group two treated with tetraclozan, group three treated with triclabendazole, and group four were left untreated. Faecal egg count was determined at day 0 before the commencement of trial and after 14, 21, 28 days of treatment. Additionally, to see worm burden 5 animals were selected (the 40 positive sheep) one sheep selected from each experimental group, and slaughtered the sheep to observe the worm recovery or not.

Anthelmintic Resistance Test

Fecal egg count (FEC) test: Efficacy of albendazole, tetraclozan and triclabendazole was estimated based on faecal egg count test within treated versus un-treated which controlled groups of sheep. The sheep were grouped into treated and untreated groups and compared the treatment group within the untreated control group. The followed equation was articulated to estimate the effectiveness of the anthelmintics [16].

$$Efficacy(\%) = \frac{MeanFECinuntreated - MeanFEC\,\mathrm{int}\,reated}{MeanFECinuntreated} \times 100$$

Fecal egg count reduction test (FECRT): This test was performed at a day 0 and at a day 14, 21, 28 when the faecal sample collected before and after treatment. Fecal egg count reduction was ≥95% indicted that effectiveness, while less than 95% indicates resistance, and also percentage of this test assessed using the formula recommended by [16].

$$\%FECRT = \frac{EPG\,\mathrm{Pr}e - Treatment - EPG14,21,28daysPost - treatment}{EPG(\mathrm{Pr}e - Treatment)} \times 100$$

OR

$$= 1 - \frac{EPG14,21,28daysPost - treatment}{EPG(\mathrm{Pr}e - Treatment)} \times 100$$

Dataset Management and Analysis

All data have been analyzed with a statistical computer package for social sciences (SPSS) version 23 software. A one way ANOVA was used to assess significance difference among treated and control group. Arithmetic means of pre and post-treatment FEC used to calculate drug efficacy [16]. Fecal egg count reduction was estimated when the efficacy ≧90% at 95% confidence interval [9]. According to [16] was reported mean FEC at a day 0 pre and post treatment at a day 14, 21, 28 was estimated when lower limit ≦94% at 95% confidence interval.

RESULT AND DISCUSSION

Field experimental study

Overall finding of the current study was revealed that the percentage of FECRT was 81.4% Triclabendazole, 63.2% Albendazole and 38.2% Tetraclozan, also efficacy of tested drugs were under standard. The arithmetic mean FEC pre-treatment was from lower 420 to higher 465 and post-treatment (14, 21 and 28 days) a ranges from lower to higher was 180 to 335, 65 to 270 and 15 to 210 respectively.

The fecal egg count reduction of this finding was revealed that the FECRT percentage and efficiency of each tested anthelmintics were 81.4% Triclabendazole, 63.2% Albendazole and 38.2% Tetraclozan. The outcome was agreed with [17] reported 56% Abendazole and 63.3% triclabendazole in Debre Birhan. On the other hand, 91.55% triclabendazole was reported in Sebeta [18], and also [19] was reported Triclabendazole 95.7% and

85.4% Albendazole in which case Triclabendazole was more effective while Albendazole not. Generally, outcome of this study, triclabendazole was effectiveness ovine Fasciola species followed by Albendazole, but tetraclozan lower effective. Therefore, the effectiveness of Triclabendazole, Albendazole and Tetraclozan below the standard, it considered that there was anthelmintic resistance in Debre Birhan district. Indeed, according to WAAVP, (1995) guidelines the faecal egg count reduction percentage is lower than 95% or the lower 95% confidence level is lower than 90% [Table 1].

The result of the current finding indicted that the average (mean± SEM) of FEC pre-treatment was 441±22.6 and post-treatment at days 14, 21, 28 was 330±27.7; 241.25 ± 29.4 and 209 ± 38.2 correspondingly. Egg pre-gram of faecal pre-treatment was a range of from 150-650 as well as post-treatment at days 14, 21, 28 was 50-700, 0-650 and 0-850 respectively.

he (mean ± SEM) faecal egg count Fasciolosis before and after treatment:- generally, triclabendazole treated groups were decreasing the number of FEC after at 14, 21 and 28 days post-treatment and some sheep were found free liver fluke after at 28 days post- treatment. Nevertheless, albendazole and tetraclozan treated group were not found free from liver fluke. Furthermore, triclabendazole treated group recover from immature and mature liver flukes [Table 2].

One way ANOVA to compare FECRT between and with grouping: - the result showed, sum squares & mean squares of the faecal egg count between treated and non-treated was statistically significantly (p=0.000). On the other hand, sum squares and mean squares among non-treated group was no statistically significance (p=0.926) [Table 3].

Table 1: Fecal egg count reduction percentage of ovine Fasciolosis at Debre Birhan districts

Measurements of the FEC	Treatment groups			
	TCBZ	ABZ	TR	NRx
Mean EPG pretreatment	465	440	440	420
Mean EPG post-treatment (14 days)	180	305	335	500
Mean EPG post-treatment(21 days)	65	145	270	485
Mean EPG post-treatment (28 days)	15	35	210	575
Efficiency (FECRT %)	81.4%	63.2%	38.2%	...

Table 2: Mean ± SEM of ovine Fasciola SPP FEC pre/post-treatment at Debre Birhan district

Measurements (EPG)	Treatment groups				
	TCBZ	ABZ	TR	NRx	Total
M. pre-treatment ±SEM)	465±48.3	440±45.8	440±51.5	420±41	441±22.6
M.post-treatment ±SEM) 14 days	180±30.9	305±47.4	335±55.8	500±35	330±27.7
M.post-treatment ±SEM) 21 days	65±16.8	145±35.3	270±37.4	485±29.9	241±29.4
M.post-treatment ±SEM) 28 days	15±7.6	35±13	210±28.7	575±43	209±38.2
Min/ Max post-treatment	200-650	150-650	200-650	200-650	.....
Min/Max post-treatment 14 days	50-350	50-500	100-550	300-700	.....
Min/Max post-treatment 14 days	0-150	0-300	100-400	350-650	.....
Min/Max post-treatment 14 days	0-50	0-100	100-300	350-850	.....

Table 3: The ANOVAs result the FECRT Fasciola among/within group at Debre Birhan district

Measurement	Group	Sum square	Mean square	p- value
Pre–treatment a day 0	Between	10187.500	3395.833	0.926
	Within	788250.000	21923.611	
Post–treatment after 14 days	Between	520500.000	173500.000	0.000***
	Within	678500.000	18847.222	
Post–treatment after 21 days	Between	1005687.500	335229.167	0.000***
	Within	343750.000	9548.611	
Post–treatment after 28 days	Between	2018687.500	672895.833	0.000***
	Within	260750.000	7243.056	

** indicated p≦0.01, * indicated p≦0.05, *** indicated p=0.000

Table 4: Multiple comparisons of different anthelmintics drugs in Debre Birhan district

Measurement (FEC)	Type of drug (i)	Type of drug (j)	p-value
Post-treatment after 14 days	Triclabendazole	Albendazole	0.049*
		Tetraclozan	0.016**
Post-treatment after 21 days	Albendazole	Triclabendazole	0.075
		Tetraclozan	0.007**
Post-treatment after 28 days	Tetraclozan	Albendazole	0.000***
		Triclabendazole	0.000***

** indicated p≦0.01, * indicated p≦0.05, *** indicated p=0.000, FEC= faecal egg count

A multiple comparison of the FECRT among different groups: - the finding showed that when triclabendazole as compared to albendazole and tetraclozan after 14 days FECRT was statistical significant (p=0.049 and 0.016). Albendazole compared to triclabendazole FECR was not statistical significant (p=0.075), but statistical significant with tetraclozan after 21 days post-treatment [Table 4].

CONCLUSION AND RECOMMENDATION

Generally this finding showed the faecal egg count reduction test (FECRT %) was commonly used to evaluated anthelmintics drugs resistance. Efficacy of tested drugs to treat a naturally infested sheep against Fasciolosis was Triclabendazole, Albendazole as well as Tetraclozan, their efficacy found 81.4%, 63.2% as well as 38.2% respectively.

The finding was revealed that efficacy tested flukicides below standard; therefore Fasciola developed anthelmintics resistance in the study area. However, Triclabendazole was more effective treated Fasciolosis. Current finding was the same opinion to the preceding research result and effectiveness against liver fluke Ethiopia concerned. Our finding different for other result, it was depending on the questioner survey about associated risk factors such as source, utilization, frequently, activities strategies deworming, available anthelmintic on the market and under dosing administration of anthelmintics drugs.

Recommendation

- Should be monitor and evaluate the application of flukic ides, avoided under dosing administration, frequency of treatments and unnecessary use of anthelmintics drugs.

- North Showa zone livestock office plus Debre Birhan livestock department should be implementing strategic control plan and extension activities avoiding drugs resistance.
- Should be additional assessment concerning under dosing, frequency of treatments and unnecessary use of anthelmintics.
- Should be further research regarding composition, national and regional anthelmintics drugs using strategically.

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1. CSA, (Central Statistical Agency). Agricultural sample survey was report on livestock and livestock characteristics. Addis Ababa. Ethiopia. 2017; 2.
2. Mas-Coma S, Bargues MD, Valero MA. Fascioliasis and other plant borne trematode zoonosis. Int J Parasitol. 2005; 35: 1255-1278.
3. Bitew M, Ibrahim N, Abdela N. Study on the prevalence of ovine Fasciolosis in and around Dawa-Cheffa, Kemissie. African Journal of Agricultural Research. 2010; 5: 2981-2985.
4. Mungube EO, Bauni SM, Tenhagen BA, Wamae LW, Nginyi JM, Mugambi JM. Prevalence and economic significance of Fasciola gigantica and hepatica in slaughtered animals in the Semi-arid Coastal Kenya. Trop Anim Health Prod. 2006; 38: 475-483.
5. Brock-well YM, Spithill TW, Anderson GR, Grillo V, Sangster NC. Comparative kinetics of serological and coproantigen ELISA and faecal egg count in cattle experimentally infected with Fasciola hepatica and following treatment with Triclabendazole. Vet Parasitol. 2013; 196: 417-426.
6. Brown DS. Fresh water snails of Africa and their medical importance. (2nd Edition.) Taylor and Francis Ltd. London. 1994; 609.
7. George MM, Paras KL, Howell SB, Kaplan RM. Utilization of composite fecal samples for detection of anthelmintic resistance in gastrointestinal nematodes of cattle. 2017; Vet Parasitol. 240: 24-29.
8. Bersissa Kumsa, Etana Debela, Megersa B. Comparative efficacy of albendazole, tetramisole and ivermectin against gastrointestinal nematodes in naturally infected goats in Ziway Oromia Regional State. Anim Vet Adv. 2010; 9: 2905-2911.
9. Boray JC. Liver fluke disease in sheep and cattle. State of New South Wales through Department of Primary Industries. 2007; 446.
10. Coles GC, Silvestre MA, Taylor JV, Prichard RK, von Samson-Himmelstjerna G, Silvestre A. The detection of anthelmintic resistance and drug tolerance in parasites veterinary importance. Vet Parasitol. 2006; 136: 167-185.
11. Tadesse Eguale, Getachew Tilahun. Molluscicidal the effects of endod phytolacca dodecandra Fasciola transmitting snails. Ethiopian Agricultural Research Organization. Institute of Pathobiology, Addis Ababa University. 2002; 25: 275-284.
12. Coles GC. Anthelmintic resistance looking to the future a UK perspective. Res Vet Sci. 2005; 78: 99-108.
13. Fair-weather I. Triclabendazole resistance treatment of immature and mature Fasciola hepatica infections in sheep. Vet Rec. 2011; 168: 514-515.
14. CSA, (Central Statistical Agency). Ethiopia agricultural sample enumeration statistical report on livestock population. Part 4. Addis Ababa, Ethiopia. 2012.
15. CSA, (Central Statistical Agency). Federal Democratic Republic of Ethiopia agricultural sample was survey report on livestock and livestock characteristics. Addis Ababa, Ethiopia. 2015; 2.
16. Coles GC, Bauer C, Borgsteede FHM, Geerts S, Klei TR, Waller PJ. World Association for the Advancement of Veterinary Parasitological methods for the detection of anthelmintic resistance in nematodes of veterinary importance. Vet Parasitol. 1992; 44: 35-44.
17. Wood AM, Amaral NK, Bairden K, Duncan JL, Malone JB, Pankavich JA, et al. World Association for Advancement of Veterinary Parasitological. 2nd edition of guidelines for the efficiency of anthelmintics in ruminants. Vet. Parasitol. 1995; 58: 181-213.
18. Skuce PJ, Zadoks RN. Liver fluke: a growing threat to UK livestock production. Cattle Pract. 2014; 21: 138-149.
19. Alvarez Sánchez MA, Mainar Jaime RC, J Perez-Garcia, Rojo-Vazquez FA. Resistance of Fasciolosis, species Fasciola hepatica to triclabendazole and albendazole in sheep in Spain. Vet Rec. 2006; 159: 424-425.